

8. (amended) The humanized polynucleotide vector according to claim 7 wherein the origin for replication is colE1 or functional portion thereof.

9. (amended) The humanized polynucleotide vector according to claim 7 wherein the origin for replication comprises a 635 base pair region of the colE1 origin of replication.

10. (amended) The humanized polynucleotide vector according to claim 1 further comprising a human-derived 3' splice sequence and a human-derived poly A sequence, both sequences located downstream of the sequence acceptance site.

11. (amended) The humanized polynucleotide vector according to claim 10 wherein the human derived 3' splice and poly A sequence are derived from human growth hormone.

C² *Sub 01* 12. (amended) A polynucleotide vector according to claim 1 wherein a 5' sequence acceptance site reads on the positive strand as GCCACCATGGCC.

13. (amended) A polynucleotide vector comprising SEQ ID No 16, SEQ ID No 27 or SEQ ID No 28.

14. (amended) A polynucleotide vector contained within a host cell deposited with the ATCC designation 98400 or ATCC designation 98401.

15. (amended) A polynucleotide vector according to claim 1 further comprising cDNA target products, and an optional internal ribosomal entry site, said cDNA target products integrated into said sequence acceptance site, said cDNA target products comprising a nucleotide sequence encoding at least one target antigen or antigenic epitope thereof alone or in combination with a nucleotide sequence encoding a cytokine or chemokine.

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16. (amended) A polynucleotide vector vaccine comprising a human derived promoter or mammalian homolog thereof which is functional in a mammalian target tissue or mammalian target cell, said promoter operably linked to a sequence acceptance site which directionally acceptance cDNA target products from rtPCR cloning via unique sites within an interrupted palindrome recognition sequence for a restriction endonuclease, an optional internal ribosomal entry site, and cDNA target products, said cDNA target products integrated into said sequence acceptance site, said cDNA target products comprising a nucleotide sequence encoding at least one target antigen or antigenic epitope thereof, and said vector lacking nucleic acid sequences encoding vector-derived polypeptides wherein, said vector lacks an antibiotic resistance encoding nucleic acid sequence.

17. (amended) A polynucleotide vector vaccine according to claim 16 wherein the target antigen is a product of a tumor associated genetic derangement.

18. (amended) A polynucleotide vector vaccine according to claim 16 wherein the target antigen is a tumor antigen, bacterial antigen, viral antigen, or parasitic antigen.

19. (amended) The polynucleotide vector vaccine according to claim 16, wherein the tumor antigen is p53, RB, ras, int-2, Hst, Tre17, BRCA-1, BRCA-2, MUC-1, HER-2/neu, truncated EGFRvIII, CEA, MyC, Myb, OB-1, OB-2, BCR/ABL, GIP, GSP, RET, ROS, FIS, SRC, TRC, WT1, DCC, NF1, FAP, MEN-1, ERB-B1 combinations thereof.

20. (amended) A polynucleotide vector vaccine according to claim 16 further comprising an additional cDNA target product comprising a nucleic acid sequence encoding a cytokine or chemokine.

21. (amended) A polynucleotide vector vaccine according to claim 20 wherein the cytokine is selected from the group consisting of interleukin 2, interleukin 3, interleukin 4, interleukin 7, interleukin 8, interleukin 12, interleukin 15, GM-CSF, tumor necrosis factor, interferon.

22. (amended) A polynucleotide vector vaccine according to claim 20 wherein the chemokine is selected from the group consisting of RANTES, MCP, MIP-E α , MIP-1 β , defensins, IP-10 and combinations thereof.

23. (amended) A method for expressing at least one target antigen or antigenic epitope thereof in cells comprising:

introducing a humanized polynucleotide vector into said cells, under conditions for expression of the target antigen or antigenic epitope thereof, said vector comprising:

a human derived promoter or mammalian homolog thereof, which is functional in said cells, said promoter operably linked to a sequence acceptance site which directionally accepts cDNA target products from rtPCR cloning via unique sites within an interrupted palindrome recognition sequence for a restriction endonuclease and,

cDNA target products, and an optional internal ribosomal entry site, said cDNA target products integrated into said sequence acceptance site, said cDNA target products comprising a nucleic acid sequence encoding at least one target antigen or antigenic epitope thereof, and said vector lacking nucleic acid sequences encoding vector-derived polypeptides, wherein said vector lacks an antibiotic resistance encoding nucleic acid sequence.

24. (amended) The method of claim 23 wherein the cells are selected from the group consisting of myocytes and professional antigen presenting cells.

25. (amended) The method of claim 23 wherein the target antigen is a tumor antigen bacterial antigen, viral antigen, or parasitic antigen.

26. (amended) The method of claim 25 wherein the tumor antigen is p53, RB, ras, int-2, Hst, Tre 17, BRCA-1, BRCA-2, MUC-1, HER-2/neu, truncated EGFRvIII, CEA, MyC, Myb, OB-1, OB-2, BCR/ABL, GIP, GSP, RET, ROS, FIS, SRC, TRC, WT1, DCC, NF1, FAB, MEN-1, ERB-B1 or combinations thereof.

27. (amended) A pharmaceutical composition comprising at least one polynucleotide vector according to claims 1, 2, 4, 5, 7 or 8-12 and a pharmaceutically acceptable carrier.

28. (amended) A pharmaceutical composition comprising the polynucleotide vector vaccine according to claims 16-21 or 22 and a pharmaceutically acceptable carrier.

29. (amended) A kit comprising the polynucleotide vector according to claims 1, 2, 4, 5, or 7-15.

30. (amended) A kit comprising the polynucleotide vector vaccine according to claims 16-22.

31 (amended) A kit according to claim 30, further comprising an expression enhancing agent.

32. (amended) The kit according to claim 31 wherein the expression enhancing agent is a mycotoxic agent.

33. (amended) The kit according to claim 32 wherein the mycotoxic agent is bupivacaine-HCl and dextrose.

34. (amended) A host cell comprising:

the polynucleotide vector of claim 16-21 or 22, wherein the host cell is capable of expressing the target antigen or antigenic epitope.

35. (amended) The host cell according to claim 34 wherein the host cell is a myocyte or professional antigen presenting cell.

36. (amended) A method of stimulating a specific immune response to at least one target antigen or antigenic epitope thereof in a mammal comprising: administration of an effective amount of a polynucleotide vector vaccine according to claim 16-21 or 22 into the mammal, said amount elicits the specific immune response to the target antigen or epitope thereof.

37. (amended) The method according to claim 36, wherein a site of administration is muscle or skin.

38. (amended) The method according to claim 36 further comprising administration of effective amount of an expression enhancing agent prior to administration of the polynucleotide vector vaccine.

39. (amended) The method according to claim 38 wherein the expression enhancing agent is a mycotoxic agent.

40. (amended) The method according to claim 39 wherein the mycotoxic agent is bupivacaine-HCl or dextrose.

41. (amended) The method according to claim 36-39 or 40 wherein the target antigen is a tumor antigen, bacterial antigen, viral antigen or parasitic antigen.

42. (amended) The method according to claim 41 wherein the tumor antigen is selected from the group consisting of P53, RB, ras, int-2, Hst, Tre 17, BRCA-1, BRCA-2, MUC-1, HER-2/neu, truncated EGFRvIII, CEA, MyC, Myb, OB-1, OB-2, BCR/ABL, GIP, GSP, RET, ROS, FIS, SRC, TRC, WT1, DCC, NF1, FAB, MEN-1, ERB-B1 and combinations thereof.

43. (amended) The method according to claim 42 wherein the method generates antigen specific cytotoxic lymphocytes to the tumor antigen or antigenic epitopes thereof.

44. (amended) A method of making a humanized polynucleotide vector comprising:
operably linking a human derived promoter or mammalian homolog thereof which is functional in a target tissue or target cells to a sequence acceptance site, said site directionally accepts cDNA target products from rtPCR cloning via unique sites within an interrupted palindrome recognition sequence for a restriction endonuclease, said vector lacking nucleic acid sequences encoding a vector-derived polypeptide wherein, said vector lacks an antibiotic resistance encoding nucleic acid sequence.

45. (amended) The method according to claim 44, wherein the human derived promoter is a RANTES promoter or portion thereof.

46. (amended) A isolate antibody comprising an antibody elicited in response to immunization with the polynucleotide vector vaccine according to claim 16-21 or 22, said antibody is specific for the target antigen or antigenic epitope thereof expressed by the mammalian target tissue or mammalian target cell.

47. (amended) The sequence acceptance site comprising nucleic acid sequences which accept cDNA target products from rtPCR cloning wherein the sequence acceptance site directionally accepts target sequence specific products from rtPCR cloning via unique sites within an interrupted palindrome recognition sequence for a restriction endonuclease.

48. (amended) The sequence acceptance site according to claim 47 wherein the restriction endonuclease is Bgl I.

49. (amended) The sequence acceptance site according to claim 47 or 48 wherein a 5' acceptance site reads on the positive strand as GCCACCATGGCC.

50. (amended) The sequence acceptance site according to claim 49 wherein a 3' acceptance site reads on the positive strand as GCCTTAAGGGC.

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51. (amended) The sequence acceptance site according to claim 47 wherein the site comprises the nucleotide sequence as depicted in Figure 2.

52. (amended) A use of a polynucleotide vector vaccine in the manufacture of a medicament for use in a method of stimulating a specific immune response to at least one target antigen or antigenic epitope thereof in a mammal, said method comprising:

administration of an effective amount of a polynucleotide vector vaccine according to claim 16-21 or 22 into the mammal, said amount elicits the specific immune response to the target antigen or epitope thereof.

53. (amended) A use according to claim 52, wherein a site of administration is muscle or skin.

54. (amended) A use according to claim 52 further comprising an expression enhancing agent.

55. (amended) The use according to claim 54, wherein the expression enhancing agent is a mycotoxic agent.

56. (amended) The use according to claim 55, wherein the mycotoxic agent is a bupivacaine-HCl or dextrose.

57. (amended) The use according to claim 52 wherein the target antigen is a tumor antigen, bacterial antigen, viral antigen or parasitic antigen.

58. (amended) The use according to claim 57, wherein the tumor antigen is selected from the group consisting of p53, RB, ras, int1-2, Hst, Tre 17, BRCA-1, BRCA-2, MUC-1, HER-2/neu, truncated EGFRvIII, CEA, MyC, Myb, OB-1, OB-2, BCR/ABL, GIP, GSP, RET, ROS, FIS, SRC, TRC, WT1, DCC, NF1, FAB, MEN-1, ERB-B1 and combinations thereof.

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59. (amended) The use according to claim 58, wherein the method generates antigen specific cytotoxic lymphocytes to the tumor antigen or antigenic epitopes thereof.

60. (amended) The humanized polynucleotide vector according to claims 1, 2, 4, 5 or 7-15, wherein the recognition sequence is recognized by BglI restriction endonuclease.

61. (amended) The humanized polynucleotide vector according to claim 7, wherein the nucleic acid sequence which allows for selection is a suppressor tRNA gene, a synthetic SupF complementation tRNA gene, or functional derivatives thereof.

C³ 62. (amended) The humanized polynucleotide vector according to claim 61, wherein the nucleic acid sequence is selected from the group consisting of SupE, SupP, SupD, SupU, SupF, SupZ, glyT, glyU, SerP, psu1⁺, psu2⁺-C34, psu3⁺ AM and psu3⁻OC.

Sub 03 63. (amended) A polynucleotide vector according to claims 1, 2, 4, 5 or 7-11, wherein a 3' sequence acceptance site reads on the position strand as GCCTTAAGGGC.

64. (amended) The humanized polynucleotide vector according to claims 1, 2, 4, 5 or 7-11, wherein the sequence acceptance site comprises the nucleotide sequence as depicted in Figure 2.

65. (amended) The method according to any of claims 23-25 or 26 wherein the method is *ex vivo*.

REMARKS

The Examiner has indicated that a Sequence listing is required for the instant application. Applicants respectfully point out that a Sequence list was originally submitted for this application on May 1, 2000 and was received by the Patent Office on May 5, 2000. On August 14, 2001, pursuant to a telephone request by the Examiner, applicants submitted by Hand Carry another copy of this Sequence list. For the Examiner's convenience, applicants again provide a